Appendix I—MARKED UP VERSION

Appendix I
Marked-Up Version of Claims
Response to 03/14/02 Office Action
13761-7001 – Due September 14, 2002

In The Claims

14. (Amended) The method of claim ~~13~~ 21 ~~further~~ comprising contacting ~~the subject's~~ a sample of the subject's nucleic acid comprising the MnSOD gene with a probe or primer which can hybridizes ~~to the polymorphic~~ a region of the MnSOD gene encoding the MTS~~mitochondrial targeting sequence of the MnSOD gene~~, said ~~polymorphic~~ region including nucleotide 351 of SEQ ID NO:1.

15. (Amended) The method of claim ~~13~~31, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS~~the identity of the allelic variant~~ comprises determining the identity of at least one nucleotide of the ~~polymorphic~~ region encoding the MTS.

16. (Amended) The method of claim 31~~13~~, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS~~the identity of the allelic variant~~ comprises performing a restriction enzyme site analysis.

17. (Amended) The method of claim ~~13~~31, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS comprises performing a~~the identity of the allelic variant is carried out by~~ single-stranded conformation polymorphism analysis.

18. (Amended) The method of ~~claim13~~claim 31, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS comprises performing an~~the identity of the allelic variant is carried out by~~ allele specific hybridization.

19. (Amended) The method of claim ~~13~~31, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS comprises performing a~~the identity of the allelic variant is carried out by~~ primer specific extension.

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20. (Amended) The method of claim ~~13~~31, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS comprises performing~~the identity of the allelic variant is carried out by~~ an oligonucleotide ligation assay.

21. (Amended) The method of claim ~~13~~31, wherein the MnSOD gene is a human MnSOD gene.

22. (Amended) The method of claim ~~13~~14, wherein the probe or primer has a nucleotide sequence from about 15 to about 30 nucleotides.

23. (Amended) The method of claim ~~13~~31, wherein the probe or primer is labeled.

24. (Amended) ~~A method for determining risk of colorectal cancer in a subject, comprising the steps of:~~

~~a.~~The method of claim 21, wherein determining whether an allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS comprises determining the base identity of a portion of genomic DNA from ~~the subject's cell~~ a sample from the subject, said genomic DNA comprising an MnSOD gene comprising a coding region for the mitochondrial targeting sequence, said portion corresponding to position 351 as defined in SEQ ID NO:1 of said MnSOD gene in said mitochondrial targeting sequence~~; and~~

~~b. correlating said base identity with a risk for colorectal cancer~~.

25. (Amended) The method of claim 24; wherein the base identity of ~~position 351~~ said portion is determined by sequencing ~~a portion of said mitochondrial targeting sequence of said MnSOD gene containing said position 351~~.

26. (Amended) The method of claim 24; wherein the base identity of said portion ~~position 351~~ is determined by digesting said portion ~~of the mitochondrial targeting sequence of said MnSOD gene~~ with an appropriate restriction endonuclease ~~appropriate to determine the base identity of said position 351~~.

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27. (Amended) The method of claim 24; wherein said base identity is determined by examining an RNA fraction from said subject's ~~cell~~ sample, whereby the identity of said genomic DNA at said position 351 can be determined.

28. (Amended) The method of claim 24; wherein the mutation in the coding region for the MTS resulting in a loss of α-helical structure in the MTS is ~~a risk for developing colorectal cancer is assessed to be greater than that of the unaffected relevant population when the base identity~~ a C at said position 351 ~~is homozygous for C~~.

--31. (New) A method of determining relative age-related risk of colorectal cancer in a subject susceptible thereto, comprising:

determining whether a first allele of a manganese superoxide dismutase (MnSOD) gene in the subject comprises a mutation in the coding region for the mitochondrial targeting sequence (MTS) of the MnSOD protein resulting in a loss of α-helical structure in the MTS;

determining whether a second allele of the MnSOD gene in the subject comprises a mutation in the coding region for the MTS of the MnSOD protein resulting in a loss of α-helical structure in the MTS;

assigning a lower risk of developing early onset colorectal cancer to a subject having no mutation in either the first or second allele of the MnSOD gene resulting in a loss of α-helical structure in the MTS;

assigning a higher risk of developing early onset colorectal cancer to a subject having mutations in both the first and second alleles of the MnSOD gene resulting in a loss of α-helical structure in the MTS; and

assigning an intermediate risk of developing early onset colorectal cancer to a subject having a mutation in only one of the first and second allele of the MnSOD gene resulting in a loss of α-helical structure in the MTS.—